

Increased Blood Glucose Level on Pelagic Fish as Response to Environmental Disturbances at East Coast Pangandaran, West Java

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Abstract. Pananjung Pangandaran Nature Reserve Area is one of the tourism destinations in Indonesia, which has large biodiversity. The environmental stressors such as pollutants can reduce the biodiversity of fish. Many physiological changes occur in response to environmental disturbances. One of these responses is blood glucose levels. The objective of this study was to evaluate the quality of the waters environment through the glucose levels of pelagic fish found in the study area. This research was conducted at Pangandaran Nature Reserve. The research was carried out using observational method. The research measured environmental parameters, such as water temperature, dissolved oxygen levels, and carbon dioxide concentrations and blood glucose level of pelagic fish. Five species had high glucose level such as *Lactarius lactarius* (173 mg/dl), *Carangoides Praeustus* (105.5 mg/dl), *Scomberomorus plumieri* (140 mg/dl), *Rastrelliger kanagurta* (202 mg/dl) and *Trichiurus lepturus* (136.3 mg/dl). In conclusion, the study indicated that the quality of East Pangandaran Beach Region, West Java has decreased quality based on the increased blood glucose levels on pelagic fish.

1. Introduction

The coastal area of Pangandaran is one of the tourism destinations in Indonesia which has large fishery resources and provides a major contribution towards the economic activities in West Java Province [1]. This area has been developed for various purposes such as sport, residential, maritime and aquaculture usages. Unfortunately, all activities of tourism and recreation in the marine environment and beach affect the quality of the sea-water, for example plastic, food waste, dish soap and “gray water” from camping and hydrocarbon residues from boat motors or OHV engines. Chemical and physical factors cause body reactions that may contribute to the health of individual fish. Fish and other aquatic animals are subject to a broad variety of stressors because their homeostatic mechanisms are highly dependent on prevailing conditions in their immediate surroundings. Stress causes increases in glucose level in fish. Therefore, the impacts of contamination on the aquatic biota can be evaluated through analysis of glucose level in fish obtained from the contaminated site [2]. One of type fishes that can be used as an indicator of environmental stress is pelagic fish who lives in the water column of coastal, ocean, and lake waters, but not on or near the bottom of the sea or the lake.

The response to environmental stress in fish is characterized by the stimulation of the hypothalamus, which results in the activation of the neuroendocrine system and a subsequent cascade of metabolic and physiological changes [3]. These changes enhance the tolerance of an organism to face an environmental variation or an adverse situation while maintaining a homeostasis status [4].



Under condition of stress, the body of the fish emits immediate responses recognized as primary and secondary responses. The primary response is the perception of an altered state by the central nervous system (CNS) and the release of the stress hormones, cortisol and catecholamines (adrenaline and epinephrine) into the circulation by the endocrine system [5].

Secondary responses occur as a consequence of the released stress hormones [6], causing changes in the blood and tissue chemistry, e.g. an increase of plasma glucose [7]. This entire metabolic pathway produces a burst of energy to prepare the fish for an emergency situation [8]. The blood glucose is the main source of energy and essential substrate for cell metabolism especially the brain cells in the fishes. Normal blood glucose levels of fish are 40-90 mg/dl [9]. Glucose homeostasis is regulated by multiple hormones. Of these, insulin is the only hormone that can decrease glucose concentration; thus it plays a key role in glucose metabolism [10]. The environmental disturbance can changes metabolism glucose of poikilothermic animals such as fish. At the time of stress, insulin inactivation causes the use of glucose by stopping cells, resulting in increased glucose in the bloodstream. The study about the Effect of Chromium, Zinc (II) and Sulphate Heptahydrate ($ZnSO_4 \cdot 7H_2O$) on Glucose Content of Freshwater Fish, *Heteropneustes fossilis* showed that the values of blood glucose have a positive correlation with concentrations of this metal. The glucose levels were found higher in the exposed fish *Heteropneustes fossilis* when compared to the control [11]. Since stress has been reported to elevate glucose levels in fishes, so the quality of the waters environment can be evaluated through the glucose levels [12]. The objective of this study was to evaluate the quality of seawater through the glucose levels in the blood of pelagic fish at East Coast Pangandaran, West Java.

2. Materials and Methods

The materials used in this study were sea water of east coast of Pangandaran, the blood of pelagic fish, amylum, O_2 Reagent, Concentrated H_2SO_4 , $MnSO_4$, and Na-thiosulfate. The tools used in this research were surgical tool, stationery, tray, burette, bucket, measuring cylinder, glucometer, glucose strip, universal pH indicator, fishing net (fishing rod), syringe, camera, erlenmeyer glass, volume pipette, plastic sample, refractometer, gloves, mercury thermometer, stative, tissue, and voinject.

This study used a quantitative descriptive approach observational method. Sampling technique was used to collect pelagic fish samples on 7 to 13 May 2017. They were taken from different sites along the east coast of Pangandaran. Sampling was primarily conducted with a 30 ft. seine net. Immediately after collecting, fishes were put into a pan of sea water for identification and counting. Blood glucose was quantified following the method of Stefani et al. (2010) where in a drop of fresh blood from caudal severed fish was touched with glucose strip inserted in a standard glucometer (Apex Biotechnology Corp.) [10] The result was obtained *in situ*. Information about fishes and habitat for all sites were collected. Habitat quality for each site was assessed according to a number of physical and water quality criteria established by Plafkin et al. [25]. Data on blood glucose was presented as mean \pm standard deviation and analysis of the data was carried descriptively out.

3. Results and Discussion

3.1. The quality of physical and chemical waters at East Coast Pangandaran

The physical and chemical of seawaters at the east coast of Pangandaran was analyzed. Temperature, salinity and pH were measured *in situ* while dissolved oxygen and BOD were analysed in the laboratory. The analysis results of physical and chemical seawater can be observed in **Table 1**.

Based on seawater quality standard criteria listed in Regulation No. 82/2001 [12], the water temperature ($28^{\circ}C$), salinity (33‰), and pH (6) in east coast of Pangandaran was definitely supportive for life of aquatic biota. Meanwhile, the data of BOD (3.8 ppm) and DO (4.5 mg/dl), were recorded in the observation location were less than the standard criteria. According to Tono et al (2010) and Tini et al (2012), the DO levels that are suitable for marine biota is greater than 5 mg/L while for BOD levels should be ≥ 20 ppm [13-14]. Thus, the status of the seawater quality in the east coast of Pangandaran belongs to a moderately contaminated category.

Table 1. The physical and chemical conditions of the sea-water in the East Pangandaran Beach

No.	Parameters	Range
1	Salinity	33‰
2	Air temperature	28°C
3	Water temperature	29°C
4	pH	6
5	Dissolved Oxygen	4.5 mg/dl
6	BOD	3.8 ppm

The temperature in this area includes a good range for fish life (25-31⁰C) [15]. The water temperature affects the rate of metabolism in the body of the fish, higher temperature can increase the metabolic rate. Water temperatures can also have a negative effect directly to the aquatic biota, the temperature affects the solubility of oxygen in the waters. The higher the temperature will lead to a lower oxygen solubility rate [16]. According to Sastrawijaya et al (2010), Solids concentration suspended in sea-waters comprising phytoplankton, zooplankton, mud, waste human, animal waste, plant waste and industrial waste, so that more concentration component in the water can increase the value of suspended solids (Total Suspended Solid). Suspended solids in east coast Pangandaran come from human waste, fishery waste and residual plants or waste from the community. Suspended solids also come from runoff land that comes from land due to rain.

Based on the quality standard according to the Regulation No. 82/2001 on the Management of Water Quality and Water Pollution Control, the pH value in the research area, belongs to the category good enough for fish life ranging from 6-9 [16]. However, the high BOD indicates that the amount of oxygen needed by microorganisms to oxidize organic matter in the water is high; it means that the water is already in a deficit of oxygen. Meanwhile, the high number of microorganisms multiplies in the water due to the abundance of food available (organic matter). Therefore, BOD always indirectly associated with levels of organic matter in the water. This is supported by the discovery of a lot of rubbish in the surrounding area. Two types of waste (organic and non-organic) can be found on the East Coast Pangandaran. The most common of organic waste are macro algae, sea grass, coconut, and paper, while non-organic waste that is found in the study area is baby diapers, plastic, food waste, and waste. Macro algae and sea grasses are two organisms stranded in the sea because the roots cannot with stand the waves. Meanwhile, paper and coconut found on the east coast of Pangandaran are delivered by the activities of coconut sellers and stalls on the beach. While, inorganic wastes (baby diapers, plastics, food scraps, and cigarettes) could be delivered from the activities of household, visitors, fishermen, and traders. The entry of domestic wastes into the waters will affect physical and chemical conditions of the water.

The garbage is heavily stagnant on the beach and some are drowned in the bottom such as baby's diapers. This can affect the physical condition of the seawater such as decreasing the value of DO and increasing the need for O₂. Domestic waste usually contains high organic matter, which will cause the decrease of dissolved oxygen so that it can cause respiration of disturbed fishes [18]. If dissolved oxygen is unbalanced, then the fish will experience stress and may cause death. This is because the body's tissue cannot bind the dissolved oxygen in the blood and the brain does not get enough oxygen supply, a common condition called anoxia. Low dissolved oxygen can lead to the presence of toxic gases such as ammonia. High levels of ammonia can cause tissue damage to the gills, where the gill plates to swell so that the respiratory system will be disrupted [15].

3.2. The morphologic of fish obtained in east coast Pangandaran

The fish are one of organisms that can be used to measure environmental degradation for some reason. First, fishes are sensitive to the wide array of direct stresses. Second, fishes integrate the adverse effect of complex and varied stresses on other components of the aquatic ecosystem, such as

habitat and macro invertebrate, by virtue of their dependence on those components for reproduction, survival and growth [19]. Only fish over 10 cm standard length were used for this study as a sample. A total of 10 species belonging to 7 families were captured, namely *Megalaspsis cordyla*, *Sardinella lemuru*, *Scomberomorus plumieri*, *Rastrelliger kanagurta*, *Lactarius lactarius*, *Carangoides praeustus*, *Pomadasys maculatus*, *Leiognathus equulus*, *Trichiurus lepturus*. and *Decapterus macarellus*. The morphology of fishes is presented in **Figure 1**.



Megalaspsis cordyla



Rastrelliger kanagurta



Sardinella lemuru's



Scomberomorus plumieri's



Lactarius lactarius



Decapterus macarellus



Pomadasys maculatus



Carangoides praeustus



Leiognathus equulus



Trichiurus lepturus

Figure 1. The morphology of fishes found in the study area.

3.3. The blood glucose level of fish

Environmental stressors such as contaminants can cause a variety of biological responses in fish ranging from the biomolecular and biochemical to population and community-level effects. To assess fishes's health, the bioindicators technique utilizes a suite of biological responses both as integrators of stress effects and as sensitive response (early-warning) indicators of existing and past environmental conditions. The growth in fish as influenced by principal ecological factors in the environment, such as seawater environment they inhabit, competition and food availability [20]. Based on measurements that have been done using a glucometer, the level of blood glucose is presented in **Table 2**.

Table 2. Blood glucose levels, weight, and length of pelagic fish

No	Species	Blood glucose level (mg/dl)	Total weight (g)	Total length (cm)
1	<i>Megalapsis cordyla</i>	51 ± 1.15	88,5	22,7
2	<i>Decapterus macarellus</i>	72.5 ± 1.73	50	15
3	<i>Scomberomorus plumieri</i>	160 ± 2.74	226	30
4	<i>Sardinella lemuru</i>	77.5 ± 0.58	36,5	16
5	<i>Restralliger kanagurta</i>	202 ± 3.26	85	17
6	<i>Lactarius lactarius</i>	201 ± 2.56	51	15
7	<i>Carangoides praeustus</i>	146 ± 1.76	54	18
8	<i>Lutjanus rivulatus</i>	94 ± 1.26	42	14,5
9	<i>Leiognathus equulus</i>	46 ± 0.96	31	13,5
10	<i>Trichiurus lepturus</i>	136.3 ± 1.69	76	46

Our study found six species with hyperglycaemic condition namely, *R.kanagurta* (202 mg/dl), *L. lactarius* (201 mg/dl), *S. Plumieri* (160 mg/dl), *C. Praeustus* (146 mg/dl), *T. lepturus* (136,3 mg/dl), and *L. rivulatus* (94 mg/dl). According to Patrice [21], normal blood glucose level of fish is 40-90 mg/dl. According to Barton (2002), the physical size of the fish determines blood glucose levels, the larger of fish body size requires more energy to support their life and as a result, equally higher secretion of glucose. Hyperglycaemia condition is not only influenced by the total length or weight but also depends on the environmental conditions and habitat quality.

In east coast Pangandaran was found domestic waste that comes from household waste such as urine, faeces, and garbage. In addition, wastes from aquaculture and agriculture were found. The entry of domestic waste into the waters will affect the physical and chemical conditions of the seawater. According to Effendi [18], domestic waste usually contains organic matter that would cause a decline in dissolved oxygen that can finally generate disturbed fish respiration. Low dissolved oxygen can cause any toxic gases such as ammonia gas. High levels of ammonia can cause damage to gill tissue, where gill plates swell so that the respiratory system will be disrupted [16]. The effect of suspended solids on fish is directly on the gills of the fish [22] and therefore oxygen which enters the body is reduced due to gills that are covered in solids. This can interfere the process of respiration so that fish need more high energy to survive. Suspended Solid is one of stress factor in fish on the east coast of Pangandaran beside ammonia; organic chemicals (demand oxygen) and low dissolved oxygen.

Many physiological changes that occur in response to environmental disturbances are now used routinely for assessing stressed states in fish. Stress responses are mediated through neuronal and endocrine pathways, known as the primary response, following initial perception of the stressor. The response to stress in fish is characterized by the stimulation of the hypothalamus, which results in the activation of the neuroendocrine system and a subsequent cascade of metabolic and physiological changes [3]. These changes enhance the tolerance of an organism to face an environmental variation or an adverse situation while maintaining a homeostasis status [24].

Under conditions of stress, the body of the fish emits immediate responses recognized as primary and secondary responses. The primary responsibility is the perception of an altered state by the central nervous system (CNS) and the release of the stress hormones, cortisol and catecholamines (adrenaline and epinephrine) into the bloodstream by the endocrine system. Secondary responses occur as a consequence of the released stress hormones [4], causing changes in the blood and tissue chemistry, e.g. an increase of plasma glucose [7]. This entire metabolic pathway produces a burst of energy to prepare the fish for an emergency [8]. Normally, the formed glucose will enter the cell will stimulate glycogenesis and lipogenesis. In the stress condition occurs response of receptors that have received stimulation, then the response will be forwarded to the hypothalamus and secrete catecholamine hormone that allows glucose into the cell. In hyperglycaemic conditions insulin inactivation occurs so that the process of glycogen formation is inhibited, glucose cannot enter the cells and cause high blood glucose levels. The rise in blood glucose is primarily generated by cortisol-mediated gluconeogenesis that also inhibits cellular uptake of circulating glucose thus increasing the levels in blood circulation. The increased level of glucose is a manifestation for the higher needs of tissues to fuel the metabolic needs of osmoregulation and an important source of energy for maintaining homeostasis in fish during chronic stress [24]. Thereby, blood glucose levels can be used as an indicator of environmental stresses [19]. In conclusion, the increase in level blood glucose in fish indicates that seawater, where were fish inhabit, were contaminated by pollutants.

4. Conclusion

Based on the results of this study can be concluded that the quality sea water of East Coast Pangandaran, West Java moderately contaminated category and the elevated level of blood glucose level on pelagic fish as a response to environmental disturbances.

5. References

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